[2] See, e.g., Mitra, S. K., *The Upper Atmosphere*, ch. III, The Royal Asiatic Society of Bengal, 1948.

[3] See, e.g., Mitra, S. K., *loc. cit.*, p. 6.

[4] *Mathematical Theory of Heat Conduction*, Ginn and Co., 1913, p. 40.

[5] Warfield, C. N., NACA, Tech. Note, No. 1200, Langley Field, Jan., 1947.

[6] *The Atmospheres of the Earth & Planets*, Univ. of Chicago Press, 1948, p. 142.

[7] *Pop. Ast.*, **55**, 322 (1947).

[8] *Pub. Univ. Tartu*, **29**, No. 5, 51 (1937).

[9] See, e.g., Brown, H., and Patterson, C., *J. Geol.*, **56**, 85 (1948).

[10] *Phys. Rev.*, **74**, 501 (1948).

[11] Reported at the June 1949 meeting of the Am. Ast. Soc.

[12] Univ. N. Mex. Pub. in Meteoritics, No. 2, 1950.

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*MULTIMOLECULAR ADSORPTION OF HORSE HEART METMYOGLOBIN ONTO FILMS OF BARIUM STEARATE**

BY ALBERT A. FISK†

GATES AND CRELLIN LABORATORIES OF CHEMISTRY, CALIFORNIA INSTITUTE OF TECHNOLOGY, PASADENA 4, CALIFORNIA‡

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In a previous paper[1] it was shown that some protein solutions, when adsorbed onto films of barium stearate, followed the classical Langmuir adsorption isotherm. The thicknesses of these unimolecular layers were calculated by using the Langmuir equation, and it was shown that these values for human and horse hemoglobin and for bovine serum albumin agreed with values in the literature for the thicknesses of these molecules.

It was felt advisable to test this technique on another protein of size and molecular weight different from the above proteins. Myoglobin was chosen because its dimensions have been recently determined by x-ray diffraction.[2]

Experimental.—The technique was adequately described in the previous paper. The thicknesses of the metmyoglobin layers adsorbed onto metallic slides covered with an optical gauge of barium stearate were determined on the ellipsometer, an optical instrument which measures the ellipticity of polarized light reflected from a metallic surface.

The metmyoglobin solutions were prepared from fresh horse heart by repeated precipitations from strong phosphate buffers. The Beckman spectrophotometer measurement of a carbonmonoxylated solution did not show the presence of any contaminating hemoglobin. The electrophoresis of a metmyoglobin solution showed that two components were present, the smaller component representing 10% of the total area. An electrophoresis of a carbonmonoxymyoglobin solution showed that the smaller component was present in one-half its concentration in the metmyoglobin

preparation. A careful separation by electrophoresis was made on a metmyoglobin solution, but subsequent electrophoreses of the separated main component revealed that the same smaller component was still present in 10% concentration. It was, therefore, assumed that the smaller component was probably not an impurity, but was metmyoglobin in the dimer or higher form. The adsorption experiments were done on this fraction at pH 6.8 in phosphate buffer of ionic strength 0.1.

Results.—The results may be seen in figure 1. The thicknesses in figure 1 are not equivalent barium stearate thicknesses, as in the previous paper, for refractive index corrections due to the relative humidity of the laboratory and the assumed method of packing of the adsorbed myoglobin

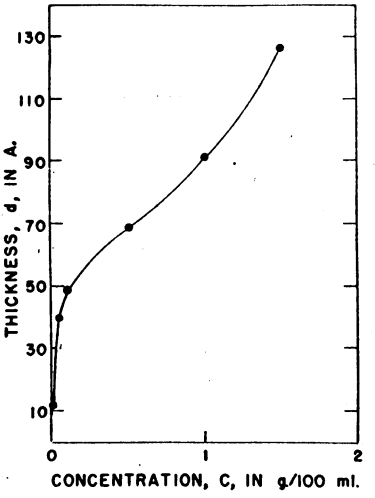

FIGURE I
ADSORPTION ISOTHERM FOR HORSE HEART METMYOGLOBIN ADSORBED ONTO BARIUM STEARATE
t = 16-19° C

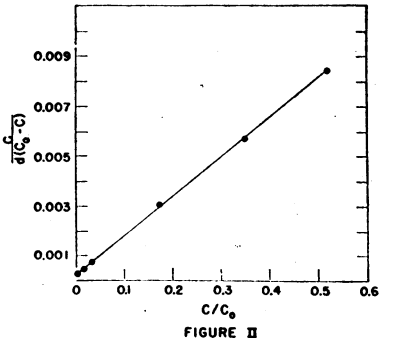

FIGURE II
B.E.T. PLOT FOR HORSE HEART METMYOGLOBIN ADSORBED ONTO BARIUM STEARATE
C_0 = 2.90 g/100 ml.

molecules led to small increases in the thicknesses. It was assumed, on the basis of x-ray diffraction data[2] and the preliminary results, that the packing model would be best approximated by assuming the closest packing of elliptical cylinders lying on their sides, the voids being filled with air. The thickness corrections were calculated by using the derivative of the Drude equation,[3]

$$\frac{dl}{l} = \frac{2dn}{n(n^2 - 1)},$$

where

l = equivalent thickness of barium stearate,

n = refractive index of barium stearate.

Interpretation of Results.—The results as shown in figure 1 indicate multimolecular adsorption of metmyoglobin onto barium stearate. The Brunauer, Emmett and Teller equation[4] for multimolecular adsorption is

$$\frac{C}{d(C_0 - C)} = \frac{1}{d_m b} + \left(\frac{b-1}{d_m b}\right)\frac{C}{C_0},$$

where

C = concentration of metmyoglobin solution,
d = calculated thickness,
C_0 = constant, the concentration at infinite thickness, assumed to be 2.90 g./100 ml.,
b = constant related to the heat of adsorption, and
d_m = thickness of a unimolecular layer of metmyoglobin molecules.

It is seen in figure 2 that a plot of $\frac{C}{d(C_0 - C)}$ against C/C_0, using the data in figure 1, gives a straight line. From the calculated slope and intercept, the thickness of a unimolecular layer of metmyoglobin molecules, d_m, was determined to be 62 A.

This value is to be compared with the maximum probable value, 57 A., for the diameter of the myoglobin molecule and the value, 9 A., for the thickness found by Kendrew[2] in his x-ray diffraction study. It would seem, therefore, that the metmyoglobin molecule has been adsorbed onto the barium stearate surface so that a long dimension, not the thickness, is perpendicular to the surface. This case is to be contrasted with the previous work[1] on hemoglobins and bovine serum albumin, where the short dimension was perpendicular to the surface. However, since myoglobin is known to have but one heme and since this heme is presumed to be attached to the side of the molecule,[2] it is not surprising to find that myoglobin may be adsorbed with the heme in a preferred orientation, either against the barium stearate surface or away from it.

The multimolecular adsorption of myoglobin, in contrast to the unimolecular adsorption of the other protein molecules studied, is not entirely unexpected in view of the fact that Taylor[5] found a considerable amount of heme-heme interaction in magnetic susceptibility measurements of myoglobin solutions. The ability of myoglobin molecules to associate was also apparent in the aforementioned preparative electrophoreses.

From the above experiment, it is apparent that the determination of protein dimensions from measurements of thin films of proteins adsorbed onto a barium stearate surface is not limited to molecules in the hemoglobin-albumin class. However, it is equally apparent that the interpreta-

tion of the results may depend on other information concerning the shape and dimensions of the molecule.

Summary.—The thickness of a unimolecular layer of horse heart metmyoglobin molecules adsorbed onto barium stearate was calculated to be 62 A. by means of the B.E.T. equation for multimolecular adsorption.

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†American Cancer Society Fellow. Present address: Biochemical Laboratory, University of Virginia Medical School, Charlottesville, Va.

‡ Contribution No. 1486.

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A COMPARISON OF THE CONTENT OF DESOXYRIBOSENUCLEIC ACID (DNA) IN ISOLATED ANIMAL NUCLEI BY CYTOCHEMICAL AND CHEMICAL METHODS

BY CECILIE LEUCHTENBERGER, ROGER VENDRELY* AND COLETTE VENDRELY*

INSTITUTE OF PATHOLOGY, WESTERN RESERVE UNIVERSITY, CLEVELAND, OHIO

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In 1948, Boivin, Vendrely and Vendrely[1, 2] presented evidence that the amount of desoxyribosenucleic acid (DNA) in nuclei of different tissues was constant for the same animal and twice that of the sperm. These authors suggested that such a constant relationship applies to all diploid animal cells and might be an expression of the genetical equipment of the cells.

While some recent work[3–6] lends support to this idea, other studies stand in contradiction. In 1949, Mirsky and Ris[6] reported analyses on the content of DNA in nuclei of various tissues of *mammals* which showed a quite different relationship to the sperm from that reported by Boivin, Vendrely and Vendrely. According to Mirsky and Ris, in calf thymus, calf lymph nodes, beef kidney and beef liver, the amount of DNA in diploid nuclei in relation to the amount of DNA in the sperm varied from 2.5 to 3:1, in contrast to the 2:1 ratio found for the same nuclei by Boivin, Vendrely and Vendrely.